


Collection of xylem sap and phloem exudates

 Zhen-Fei Chao  Dai-Yin Chao

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 An abbreviated version of this protocol was published in Science Advances in Sep 2021

NPF transporters in synaptic-like vesicles control delivery of iron and copper to seeds

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Detailed protocol

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For collection of xylem sap, plants were grown in hydroponic culture or soil watered with Hoagland nutrient solution weekly. After the inflorescence stem is ~10 cm length (6-7 weeks for short day photoperiod and 4-5 weeks for long day photoperiod), remove all rosette leaves first, then the stems were cut using a sharp razor 1-2 cm above the basal stems. Each plant was covered with a 2 mL tube or all the plants were put in a box covered with cling film to keep a high humidity and incubated in an illuminated growth chamber and xylem sap was carefully collected using pipette during a 3-4 h period. Xylem sap collected during the first 10 min was discarded to avoid contamination from damaged cells.

Phloem exudates were collected by using the EDTA-facilitated method. Briefly, whole rosettes of 5-week-old plants grown in artificial soil were removed from the root using a sharp razor and immersed in deionized water before individual leaves were detached at the petiole. Four leaves (the 9th and 10th) collected from two plants were pooled together with the petioles put in a 1.5 mL tube filled with deionized water. The samples were then put in a box covered with cling film to keep a high humidity and incubated in an illuminated growth chamber for 15 min to flush of xylem sap. The petioles were then recut under 5 mM Na₂-EDTA (pH 7.5) under low light and were placed in final 250 µL of 5 mM Na₂-EDTA (pH 7.5) for incubation in darkness for 1 h in a high-humidity box.

How to cite: (Readers should cite both the Bio-protocol preprint and the original research article where this protocol was used)

1. Chao, Z. and Chao, D. (2021). Collection of xylem sap and phloem exudates. Bio-protocol Preprint. bio-protocol.org/prep1484.
2. Chao, Z., Wang, Y., Chen, Y., Zhang, C., Wang, P., Song, T., Liu, C., Lv, Q., Han, M., Wang, S., Yan, J., Lei, M. and Chao, D. (2021). NPF transporters in synaptic-like vesicles control delivery of iron and copper to seeds. Science Advances 7(36). DOI: [10.1126/sciadv.abh2450](https://doi.org/10.1126/sciadv.abh2450)

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